PCT WORLD INTELLECTUAL PROPE
International Bu

INTERNATIONAL APPLICATION PUBLISHED UNDER

(51) International Patent Classification 6 :
C07B 33/00, B01J 31/18

A2

(11) In

WO 9608455A2

(43) International Publication Date: 21 March 1996 (21.03.96)

(21) International Applicati n Number: PCT/US95/11522

(22) International Filing Date: 14 September 1995 (14.09.95)

(30) Priority Data:

08/306,801	15 September 1994 (15.09.94)	US
08/520,842	12 September 1995 (12.09.95)	US

(81) Designated States: CA, JP, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).

Published
Without international search report and to be republished upon receipt of that report.

(71) Applicant: ABBOTT LABORATORIES [US/US]; CHAD 0377/AP6D-2, 100 Abbott Park Road, Abbott Park, IL 60064-3500 (US).

(72) Inventors: CHORGHADE, Mukund, S.; 5048 Adele Drive, Gurnee, IL 60031 (US). DOLPHIN, David, H.; 4464 West 12th Avenue, Vancouver, British Columbia V6R 2R2 (CA). HILL, David, R.; Apartment E, 5312 George Court, Gurnee, IL 60031 (US). HINO, Fumio; 1-35-23, Noyawa, Setagaya-ku, Tokyo 154 (JP). LEE, Elaine, C.; 755-B Brookvale Drive, Wheeling, IL 60090 (US).

(74) Agents: ELDER, Richard, A. et al.; Abbott Laboratories, CHAD 0377/AP6D-2, 100 Abbott Park Road, Abbott Park, IL 60064-3500 (US).

(54) Title: USE OF SYNTHETIC METALLOPORPHYRINS FOR PREPARATION AND PREDICTION OF DRUG METABOLITES

(57) Abstract

A method for the systematic and efficient synthetic preparation and identification of metabolites of a pharmaceutical product in order to study possible toxic and/or otherwise biologically-active metabolites of such pharmaceutical products as early and conveniently as possible in the very expensive drug development process, comprising adding samples of the pharmaceutical product to a series of combinations of a synthetic metalloporphyrin (SMP) with a synthetic metalloporphyrin-co-oxidizing reagent in the presence of a suitable solvent, under specified conditions, in a manner such that each sample of pharmaceutical product is reacted with a different combination of synthetic metalloporphyrin, SMP-co-oxidizing reagent and solvent, followed by separation and isolation of the resulting oxidative products, then confirmation of the identity of metabolites from the pre-identified oxidative products by appropriate animal model studies, and subjecting actual metabolites prepared in larger quantities by the above method to toxicologic, pathologic, histopathologic, mechanistic or genotoxic ng in order to identify toxic and/or otherwise metabolically-active beneficial or detrimental individual metabolites.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AT	Austria	**GB**	United Kingdom	**MR**	Mauritania
AU	Australia	**GE**	Georgia	**MW**	Malawi
BB	Barbados	**GN**	Guinea	**NE**	Niger
BE	Belgium	**GR**	Greece	**NL**	Netherlands
BF	Burkina Faso	**HU**	Hungary	**NO**	Norway
BG	Bulgaria	**IE**	Ireland	**NZ**	New Zealand
BJ	Benin	**IT**	Italy	**PL**	Poland
BR	Brazil	**JP**	Japan	**PT**	Portugal
BY	Belarus	**KE**	Kenya	**RO**	Romania
CA	Canada	**KG**	Kyrgystan	**RU**	Russian Federation
CF	Central African Republic	**KP**	Democratic People's Republic of Korea	**SD**	Sudan
CG	Congo			**SE**	Sweden
CH	Switzerland	**KR**	Republic of Korea	**SI**	Slovenia
CI	Côte d'Ivoire	**KZ**	Kazakhstan	**SK**	Slovakia
CM	Cameroon	**LI**	Liechtenstein	**SN**	Senegal
CN	China	**LK**	Sri Lanka	**TD**	Chad
CS	Czechoslovakia	**LU**	Luxembourg	**TG**	Togo
CZ	Czech Republic	**LV**	Latvia	**TJ**	Tajikistan
DE	Germany	**MC**	Monaco	**TT**	Trinidad and Tobago
DK	Denmark	**MD**	Republic of Moldova	**UA**	Ukraine
ES	Spain	**MG**	Madagascar	**US**	United States of America
FI	Finland	**ML**	Mali	**UZ**	Uzbekistan
FR	France	**MN**	Mongolia	**VN**	Viet Nam
GA	Gabon				

USE OF SYNTHETIC METALLOPORPHYRINS FOR PREPARATION AND PREDICTION OF DRUG METABOLITES

Technical Field

Synthetic metalloporphyrin (SMP) compounds serve as models of oxidative catalysts in biological systems. Oxidative products of pharmaceutical compounds, which products are useful in the prediction of biological metabolites, may be systematically and efficiently prepared by reacting the pharmaceutical compounds with specified combinations of synthetic metalloporphyrin catalysts, SMP-co-oxidizing reagents and solvents, followed by separation and identification of such oxidative products, which oxidative products may then be subjected to toxicologic, pathologic, histopathologic, mechanistic or genotoxic testing for determining the toxicity and other biological properties of the metabolites of the original pharmaceutical compounds.

Background of the Invention

In humans and other animals most drugs are metabolized in the liver. Many drug metabolites are formed by oxidative mechanisms catalyzed primarily by heme- and cytochrome-containing enzymes. Of these, the cytochrome P450-dependent monooxygenases provide the primary catalysis in most biological oxidations (cf., Cytochrome P-450: Structure, Mechanism and Biochemistry, P.R. Ortiz de Montellano, ed., Plenum Press, NY, 1986).

The metabolic process which a drug compound undergoes in the body contibutes in large measure to the efficacy of the compound for a particular purpose (sometimes actually resulting in creation of the active compound itself), to whatever side-effects a compound may possess, and to the presence or absence of toxicity or undesirable biological activity of its metabolites. These factors are major contributors to the success or failure of a particular pharmaceutical compound and the importance of the metabolic process has been sufficient to justify the vast amounts of research effort which has been expended in the past thirty years in studying the mechanisms of the oxidative metabolic processes.

Pharmacologists, being aware of the importance of drug metabolites to the future of pharmaceutical product candidates, have been involved in the attempts to identify and isolate such compounds. They have traditionally tried to obtain sufficient quatities of these metabolites as early as possible in the very expensive drug development process, in order to conduct further toxicological and pharmacological studies on them.

Several problems are associated with the use of biological systems in studying drug metabolism, however. In particular, both animal and *in vitro* metabolic studies produce very small amounts of metabolites, thus making identification of these metabolites difficult. These metabolites generally must be isolated in order to be identified, and pharmacologists do not know in advance for which potential metabolites they should be looking. Also, animal studies are notoriously expensive to conduct, since large numbers of animals are required for these metabolic studies, and even when identified, the metabolites may not be easily or efficiently synthesized for purposes of further testing, especially when larger amounts of metabolites are required for such testing.

Recently, investigators have begun to study model systems of the biological oxidations in which synthetic metalloporphyrins are utilized as mimics of the cytochrome P450-dependent monooxygenase catalysts. A limited number of reviews of the literature in this new field have been published, including those by Xie and Dolphin ("Biological Oxidations with Heme Proteins," in Metalloporphyrins Catalyzed Oxidations, F. Montanari and L. Casella, eds., Kluwer Academic Publishers, The Netherlands, **1994**, pp 269-306); and Montanari *et al.* (*Rev. Heteroat. Chem.*, 6:94-141 (**1992**)).

The first SMPs studied were found to be unstable, but improvements in molecular stability and increases in the turnover of catalytic reactions have been obtained with the introduction of additional atoms into the synthetic metalloporphyrin molecules. The work of Dolphin and others has shown that addition of halogen atoms onto the aryl groups and the β-pyrrolic positions of meso-tetraarylporphyrins makes intermediate oxo-porphyrin complexes more electron deficient and more sterically protected and thus provides for more effective oxidation catalysis (see, for example, Xie and Dolphin, op. cit.).

However, model studies with these halogenated synthetic metalloporphyrins have been hampered by lack of convenient access to these catalytic compounds themselves. As these catalysts are not commercially available, they must generally be prepared in the researcher's laboratories. For examples of methods currently used for synthesis of synthetic metalloporphyrins, representative procedures are given by Dolphin *et al.*, U.S. Patent No. 4,892,941, issued Jan. 9, 1990; Traylor *et al., Inorg. Chem.*, 26:1338-1339 (**1987**); Rocha Gonsalves *et al., Tetrahedron Lett.*, 32:1355-1358 (**1991**); Hoffmann *et al., Tetrahedron Lett.*, 31:1991-1994 (**1990**); and Wijesekera *et al., Angew. Chem., Int. Ed. Engl.*, 29:1028-1030 (**1990**).

To date, few other uses of synthetic metalloporphyrins for the study of the oxidative metabolism of drugs have been reported, however. Carrier *et al.* (*Bull. Soc. Chim. Fr.*, 130:405-416 (**1993**)), who studied lidocaine oxidation with various cytochrome P450 model systems and produced thereby some of the known primary metabolites of lidocaine, have suggested that reaction conditions and the metalloporphyrins themselves might be varied to give differing amounts of oxidation products, or in some cases, different products entirely. (In contrast, by applying the novel method of the present invention, the remaining known metabolites, as well as some additional oxidation products which are being considered as possible additional metabolites in ongoing studies, have been produced.)

Matsumoto *et al.* (*Drug. Metab. Disp.*, 19:768-780 (**1991**)), in a very narrow study, examined the oxidation of piperidine ring systems by cytochrome P450 model metalloporphyrins. Also, novel oxidation products of erythromycin, which are not, however, biological metabolites of that compound, have been prepared by D.R. Hill *et al.* (*Tetrahedron Letters*, manuscript in preparation).

It was therefore an objective of this invention to provide pharmacologists with a method of systematically and efficiently producing and identifying the metabolites of drug candidates to permit them to determine whether these metabolites possess any unacceptable toxicity profiles and/or if they have either desirable or undesirable biological activity as early as possible in the expensive drug development process. It was also an objective of this invention to provide a method of producing and identifying oxidative products of a pharmaceutical candidate from which the metabolites of a pharmaceutical product could be identified before animal or biological studies are done.

It was another objective of this invention to provide a synthetic method of producing quantities of oxidation products of drug candidates, which products may have been identified as metabolites by biological testing, in quantities sufficient to allow for toxicological and futher biological tests thereon, at an early stage in the discovery process. It was a further objective of this invention to provide acceptable ways of reducing the amount of animal testing required in the development of a drug candidate.

Summary of the Invention

The present application describes a method for the systematic and efficient synthetic preparation and identification of metabolites of a pharmaceutical product in order to study possible toxic and/or otherwise biologically-active metabolites of such pharmaceutical products as early and conveniently as possible in the very expensive drug development process, comprising adding samples of the pharmaceutical product to a series of combinations of a synthetic metalloporphyrin (SMP) with a synthetic metalloporphyrin-co-oxidizing reagent in the presence of a suitable solvent, under specified conditions, in a manner such that each sample of pharmaceutical product is reacted with a different combination of synthetic metalloporphyrin, SMP-co-oxidizing reagent and solvent, followed by separation and isolation of the resulting oxidative products, then confirmation of the identity of metabolites from the pre-identified oxidative products by appropriate animal model studies, and subjecting the actual metabolites prepared in larger quantities by the above method to toxicologic, pathologic, histopathologic, mechanistic or genotoxic testing in order to identify toxic and/or otherwise metabolically-active beneficial or detrimental individual metabolites.

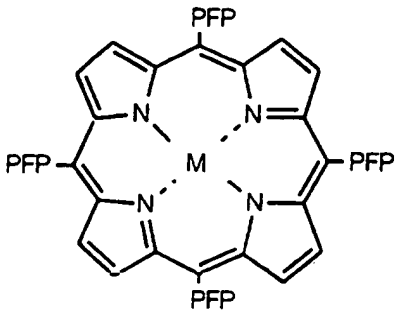
Detailed Description of the Invention

The present invention is directed to a process for the systematic preparation of oxidative products of a drug candidate compound, comprising reacting the drug candidate compound with a series of combinations of a member of group of synthetic metalloporphyrins, as defined below, with a member of a group of SMP-co-oxidizing reagents, as defined below, in the presence of a suitable solvent, such as, for example, methylene chloride, acetonitrile, acetonitrile/water, methanol/water, buffered aqueous solutions thereof, or the like, for a period of up to twenty-four (24) hours, at a temperature from 0°C to reflux temperature of the solvent, in a manner such that each sample of drug compound is reacted with a different combination of synthetic metalloporphyrin, SMP-co-oxidizing reagent and solvent, followed by separating and isolating each resulting oxidative product by gas, liquid/liquid, or solid/liquid chromatography, HPLC, or the like. Said oxidative products may then be identified by analytical means such as, for example, NMR, MS, IR, or UV spectroscopy, or the like.

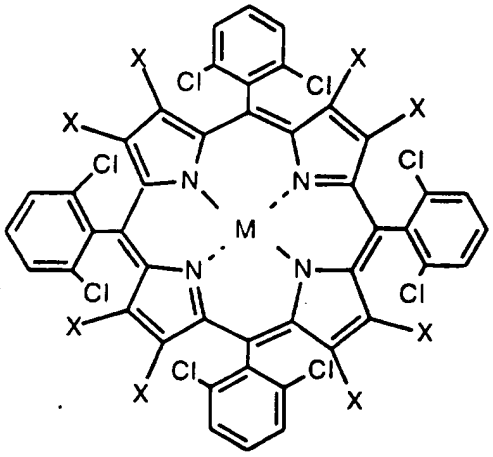
These oxidative products are then identified to a pharmacologist who uses them as predictors to identify actual metabolites of the original drug candidate

compound in studies with appropriate animal models. And the actual metabolites are then subjected (in larger quantities prepared by methods according to the above process which has been optimized to prepare these specific metabolites) to various biological tests in order to identify toxicity and/or other desirable or undesirable biological activity of these metabolites as early as possible in the very expensive drug development process.

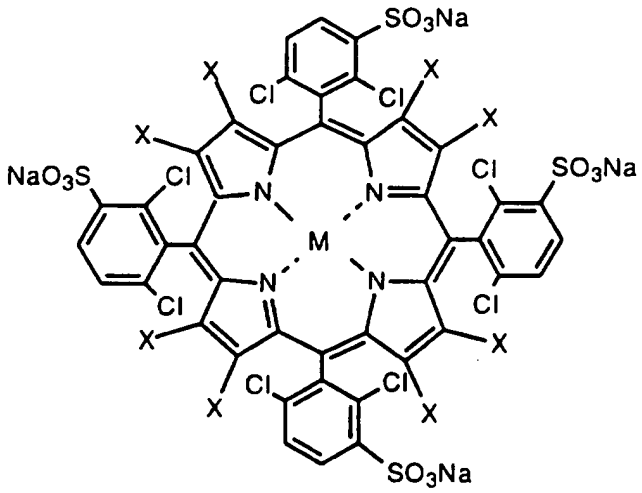
The term "synthetic metalloporphyrin," as used herein, refers to porphyrin compounds having the structures:



, (1), wherein PFP represents perfluorophenyl and M is selected from the group consisting of iron, manganese, chromium, ruthenium, cobalt, copper and nickel;



, (2), wherein X is Cl, Br, NO_2, CN or sulfonate, and M is as described above; or



, (3), wherein X and M are as described above.

The abbreviations for the synthetic metalloporphyrins used herein include: perfluorotetraphenyl Fe(III) porphyrin for the compound of formula (1) above, wherein M is iron; perfluorotetraphenyl Mn(II) porphyrin for the compound of formula (1) above, wherein M is manganese; octachloro-octabromo Fe(III) porphyrin for the compound of formula (2) above, wherein X is Br and M is iron; octachloro-octabromo Mn(II) porphyrin for the compound of formula (2) above, wherein X is Br and M is manganese; octachloro-octachloro Fe(III) porphyrin for the compound of formula (2) above, wherein X is Cl and M is iron; octachloro-octachloro Mn(II) porphyrin for the compound of formula (2) above, wherein X is Cl and M is manganese; octachloro-octabromo tetrasulfonato Fe(III) porphyrin for the compound of formula (3) above, wherein X is Br and M is iron; octachloro-octabromo tetrasulfonato Mn(II) porphyrin for the compound of formula (3) above, wherein X is Br and M is manganese; octachloro-octachloro tetrasulfonato Fe(III) porphyrin for the compound of formula (2) above, wherein X is Cl and M is iron; and octachloro-octachloro tetrasulfonato Mn(II) porphyrin for the compound of formula (2) above, wherein X is Cl and M is manganese.

Other synthetic metalloporphyrins suitable for use herein may have effective catalytic activity as a result of substitution with electron-withrawing and sterically-protecting groups, such as, for example, substitution of nitro, cyano or sulfonate for the chlorine atoms on the phenyl rings and/or the porphyrin rings of compounds of formulas (2) or (3) above, or carboxyl substitution for the sulfonato groups of compounds of formulas (2) or (3) above. In general, these synthetic metalloporphyrins are highly reactive, are not rapidly destroyed under strong oxidizing conditions, and are capable of effecting catalytic oxidations with high turnover numbers.

The preferred synthetic metalloporphyrins envisioned for use in this invention are the compounds selected from the group of compounds comprising formulas (2) or (3) above.

The more preferred synthetic metalloporphyrins for use herein are the compounds of formulas (2) or (3) above wherein M is iron or manganese, and are selected from the group comprising octachloro-octabromo Fe(III) porphyrin, octachloro-octabromo Mn(II) porphyrin, octachloro-octachloro Fe(III) porphyrin, octachloro-octachloro Mn(II) porphyrin, octachloro-octabromo tetrasulfonato Fe(III) porphyrin, octachloro-octabromo tetrasulfonato Mn(II) porphyrin, octachloro-octachloro tetrasulfonato Fe(III) porphyrin, and octachloro-octachloro tetrasulfonato Mn(II) porphyrin.

The synthetic metalloporphyrins may be prepared by known methods (see the references cited in the Background, above) wherein a suitable zinc-containing metalloporphyrin, such as *meso*-tetrakis(2,6-dihalophenyl)-porphyrinato-zinc(II), wherein "halo" is chloro, bromo, fluoro, or indo, is reacted with one of several active halogenating agents, followed by removal and replacement of the zinc atom with the desired active metal ion. They may also be prepared by an improved method for the preparation of a porphyrin-ring halogenated synthetic metalloporphyrin, wherein the halogenating agent may be a free halogen, such as Cl_2 or Br_2, in a suitable polar solvent, such as methanol, ethanol, or the like, and the reaction may be performed at lower temperatures, thus resulting in enhanced yields of the desired product.

Such a synthetic metalloporphyrin may be more preferably prepared by reacting a suitable zinc-containing metalloporphyrin, such as *meso*-tetrakis(2,6-dichlorophenyl)-porphyrinato-zinc(II), for example, with a free halogen, such as Cl_2 or Br_2, in a suitable polar solvent, such as methanol, ethanol, or the like, at a temperature of from 0°C to ambient, followed by removal and replacement of the zinc atom with the desired active metal ion.

The synthetic metalloporphyrins may be attached to support materials in adsorbed, covalently- or ionically-bonded manners, for example, adsorbed onto diatomaceous earth. Such adsorbed preparations may be utilized in the form of suspensions or in fixed format, such as, for example, in columns.

The term "SMP-co-oxidizing reagent," as used herein, refers to those oxidizing agents suitable for use with a synthetic metalloporphyrin, and include, for example, iodosobenzene, sodium hypochlorite, potassium monopersulfate, ozone, and peroxides, such as hydrogen peroxide, m-chloroperbenzoic acid, cumene hydroperoxide or *tert*-butyl hydroperoxide.

Preferred SMP-co-oxidizing reagents are those selected from the group comprising iodosobenzene, sodium hypochlorite, *tert*-butyl hydroperoxide and potassium monopersulfate.

The co-oxidizing reagent is preferably added to the reaction mixture gradually, in small quantities, with a fresh charge of oxidant being added after a period of 3 hours.

The solvent in which the above reactions are carried out may be any solvent known to those skilled in the art which does not interact unfavorably with the synthetic metalloporphyrin and/or the co-oxidizing reagent. The solvent may be selected to favor solubility of the drug compound or the synthetic metalloporphyrin, or for ease of recovery and purification of the product.

Preferred solvents are those selected from the group comprising CH_2Cl_2, CH_3CN, 20% methanol in H_2O, 20% CH_3CN in H_2O, or aqueous solutions buffered to various pH levels.

In a preferred embodiment of this process, the synthetic metalloporphyrins are selected from the group comprising octachloro-octabromo Fe(III) porphyrin, octachloro-octabromo Mn(II) porphyrin, octachloro-octachloro tetrasulfonato Fe(III) porphyrin, and octachloro-octachloro tetrasulfonato Mn(II) porphyrin; the SMP-co-oxidizing reagents are selected from the group comprising iodosobenzene, sodium hypochlorite, *tert*-butyl hydroperoxide, and potassium monopersulfate; and the solvents are those selected from the group comprising CH_2Cl_2, 20% CH_3CN in H_2O, and buffered aqueous solutions.

The combining and reacting of the pharmaceutical compound, the synthetic metalloporphyrins, SMP-co-oxidizing reagents and the solvents may be achieved either simultaneously or serially with or without appropriate automated means, including the use of robotic devices. A "kit" of metalloporphyrin reagents and oxidizers may also be prepared for convenient use of this novel process, and is considered to be within the scope of the invention.

It is intended that this invention include optimization of reaction conditions by easy, rapid and repetitive experimentation to identify the appropriate combination of solvent, metalloporphyrins, oxidant and reaction conditions that produces the maximum number and/or amount of metabolites or of one or more desired metabolites. This logically leads to a subsequent scaled-up optimal process by which large amounts of one or more desired metabolites may be produced.

"Appropriate animal models" for use in confirming that the oxidative products prepared by the process above are actually metabolites of the pharmaceutical product being studied include those identified by methods well-

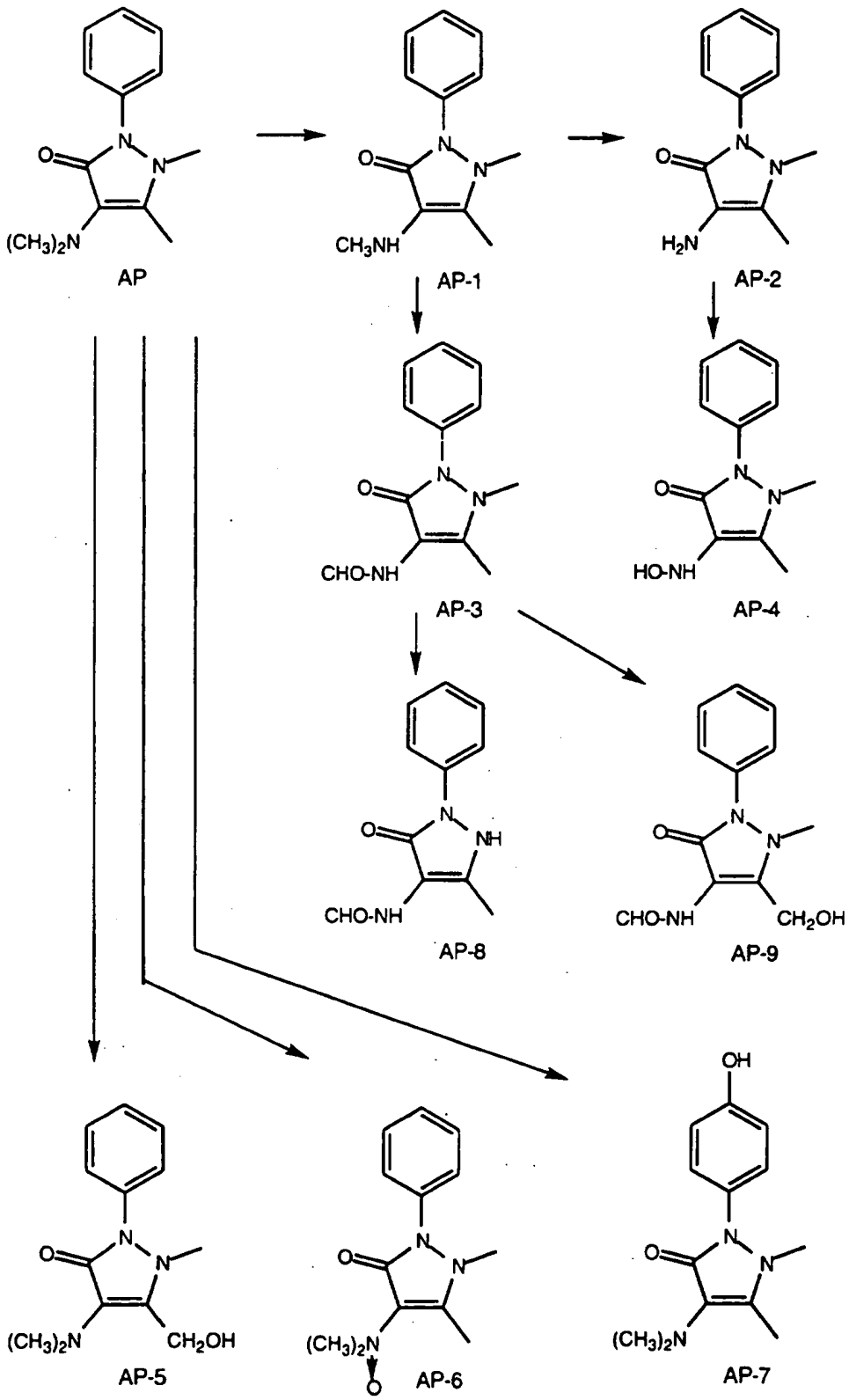
known to pharmacologists for determining animal species which have metabolic processes for the particular product category which are similar to those of humans.

The process of the invention may be used in combination with an examination of the oxidative products produced thereby in toxicity tests, such as for example, acute, sub-chronic, or chronic studies involving clinical pathologic, histopathologic, mechanistic or genotoxicity protocols, or in other screens or protocols in use for determining biological activity, for identifying toxic or metabolically-active metabolites of a drug candidate.

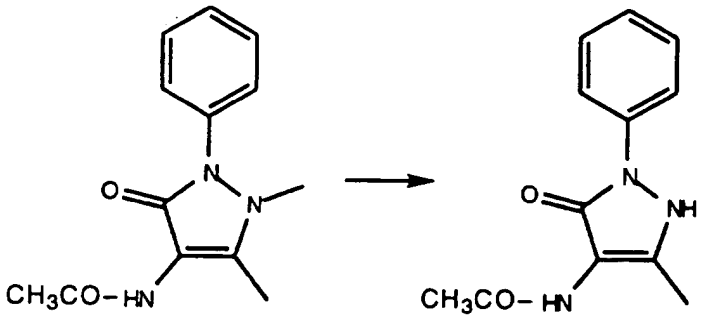
Schemes

The oxidation products of the reactions herein are illustrated in the following schemes. Scheme 1 illustrates aminopyrine and its oxidation products. Scheme 2 shows the oxidation of 4-acetylaminoantipyrine (AP-10). Scheme 3 illustrates the oxidation product of 3-hydroxymethylaminopyrine (AP-5). Scheme 4 shows lidocaine and its oxidation products. Scheme 5 illustrates dimethylaniline and its oxidation products. Scheme 6 diagrams the preparation of octachloro-octahalo Zn(II) porphyrins, wherein the octachloroporphyrin on the left is reacted with molecular chlorine or bromine in methanol to give the octachloro-octahalo-porphyrin on the right. Scheme 7 shows the oxidation products of ABT-418. Scheme 8 shows the oxidation products of odapipam.

Scheme 1

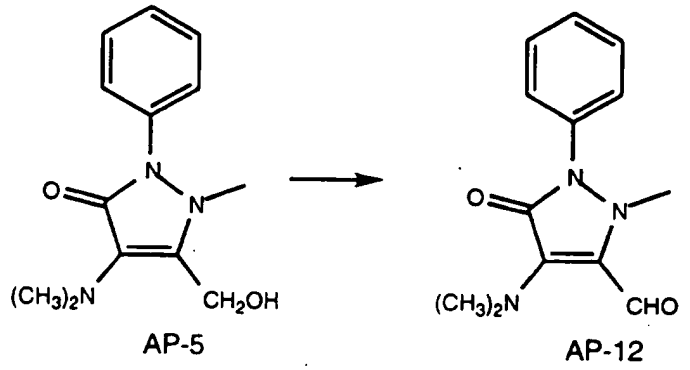


Scheme 2

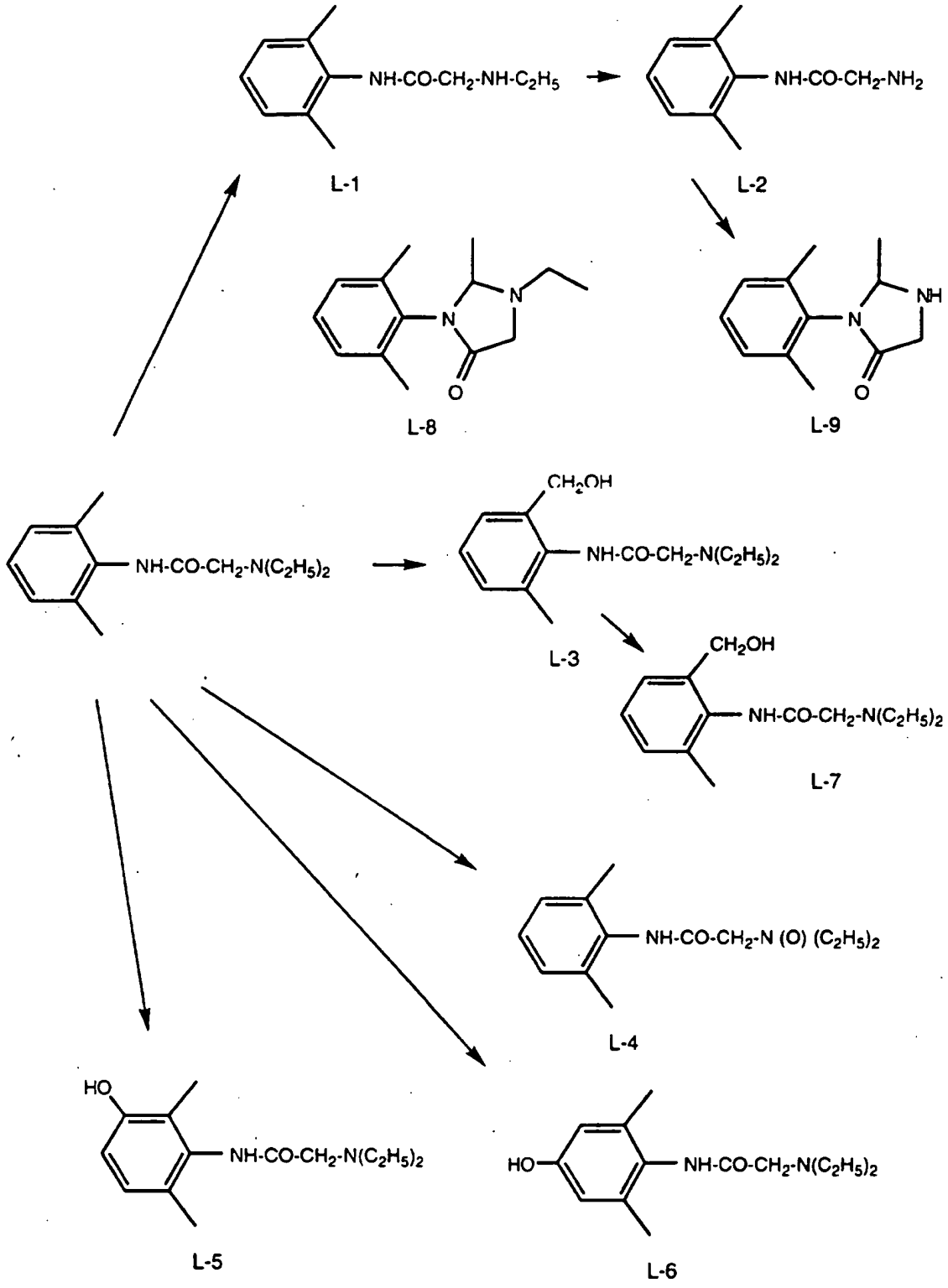


5

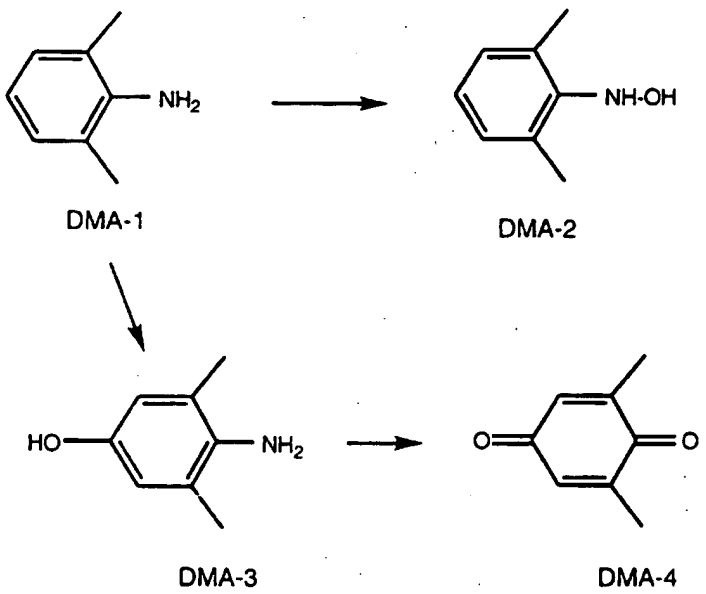
Scheme 3



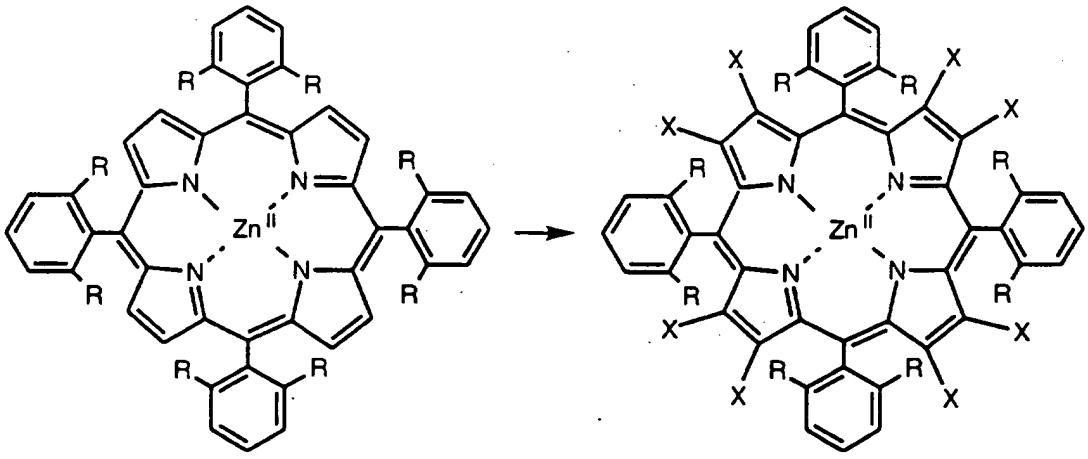
Scheme 4


NH-CO-CH2-NH-C2H5
NH-CO-CH2-NH2
L-1
L-2
L-8
L-9
NH-CO-CH2-N(C2H5)2
CH2OH
L-3
L-7
NH-CO-CH2-N (O) (C2H5)2
L-4
HO
L-5
L-6

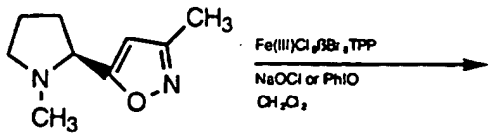
Scheme 5



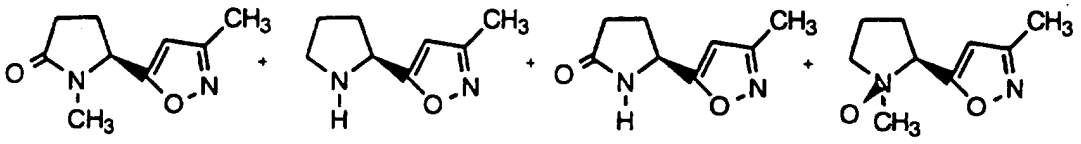
Scheme 6



Scheme 7

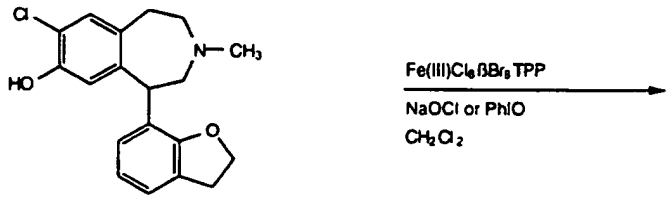


ABT-418

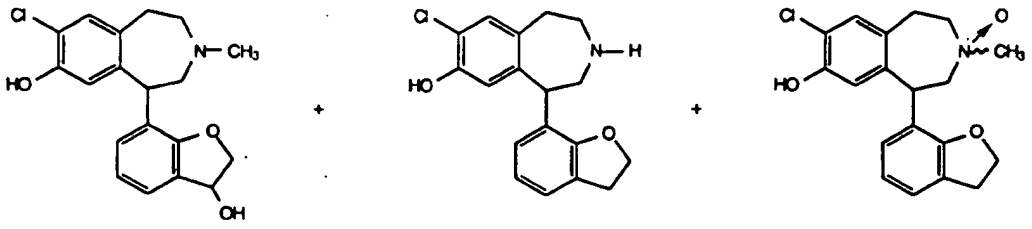


37 % 39-40 % 4-5 % < 1 %

Scheme 8



Odapipam



Cis and Trans

The following examples illustrate particular embodiments of the invention, and are not intended as a limitation upon the scope thereof.

Example 1

Aminopyrine metabolites

Aminopyrine hydrochloride was prepared from the free base (Aldrich) by treatment with HCl in ether. Iodosobenzene (1650 mg, 7.5 mmol, prepared according to the method of Saltzman *et al., Org. Synth.*, 43:60-61 (1963)) was added with stirring at room temperature in portions every 30 min to a solution of octachloro-octachloro tetrasulfonato Fe(III) porphyrin (6.2 mg 3.9 μmol) in 50 mL of 80:20 H_2O:CH_3CN containing 678 mg (3.0 mmol) of aminopyrine. Two hr after the last addition of the oxidant, the solution was evaporated under reduced pressure at 50°C. The residue was dissolved in 20 mL of aqueous Na_2CO_3 solution, and the solution was extracted with CH_2Cl_2 (3 x 50 mL). The organic layer was dried over Na_2CO_3 and evaporated under reduced pressure. The residue was chromatographed on silica gel, eluting with 1:25 methanol:methylene chloride. Seven oxidation products were obtained from the eluate (see Scheme 2): 2,3-dimethyl-4-monomethylamino-1-phenyl-3-pyrazolin-5-one (AP-1); 4-amino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one (AP-2); 4-formylamino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one (AP-3); 2,3-dimethyl-4-hydroxyamino-1-phenyl-3-pyrazolin-5-one (AP-4); 4-dimethylamino-3-hydroxymethyl-2-methyl-1-phenyl-3-pyrazolin-5-one (AP-5); 4-dimethylamino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one-4-N-oxide (AP-6); and 4-dimethyl-2,3-dimethyl-1-(4-hydroxyphenyl)-3-pyrazolin-5-one (AP-7), 4-formylamino-2-methyl-1-phenyl-3-pyrazolin-5-one (AP-8), and 4-formylamino-3-hydroxymethyl-2-methyl-1-phenyl-3-pyrazolin-5-one (AP-9).

The yields of these products were as shown in the column for Example 1 in Table 1 below. The AP-4, AP-7, AP-8 and AP-9 compounds are new metabolites, while the others are known metabolites of the oxidation of aminopyrine with liver microsomes or purified cytochrome P-450s.

Table 1. Yields of aminopyrine metabolites under varying reaction conditons

	yields of metabolites (%)		
metabolite	Example 1	Example 2	Example 3
AP-1	4		
AP-2	53		
AP-3	17		
AP-4	2		
AP-5	2	78	
AP-6	1		
AP-7	3		16
AP-8	1		
AP-9	1		

Example 2
Alternate oxidation of aminopyrine at pH 1

Iodosobenzene (66 mg, 0.3 mmol) was added with stirring at room temperature in portions over 30 min to a solution of octachloro-octachloro tetrasulfonato Fe(III) porphyrin (1 mg, 0.6 µmol) in 2 mL of 1:4 acetonitrile:pH 1 H_2O (adjusted with 0.2 N HCl) containing 28 mg (0.12 mmol) of aminopyrine. The solution was evaporated under reduced pressure. The residue was dissolved in satd aq. Na_2CO_3, and the products were extracted with 3 x 1 mL of methylene chloride. The organic layer was chromatographed over silica gel, eluting with methylene chloride. The solvent was evaporated under reduced pressure, and the residue was crystallized from methylene chloride:hexanes. Compound AP-5 was obtained as the major metabolite (22.7 mg, 78% yield), and was the only one identified quanitatively.

Example 3
Alternate oxidation of aminopyrine in organic solvents

Aminopyrine hydrochloride was prepared from the free base (Aldrich) by treatment with HCl in ether. Iodosobenzene (66 mg, 0.3 mmol) was added with stirring at room temperature in portions over 30 min to a solution of octachloro-octachloro tetrasulfonato Fe(III) porphyrin (1 mg, 0.6 µmol) in 2 mL of 1:20 methanol:CH_3CN containing 30 mg (0.13 mmol) of aminopyrine hydrochloride.

The reaction was stirred for 10 min after the last portion of oxidant was added, and the solution was evaporated under reduced pressure. The residue was chromatographed over alumina, eluting with 5:1 methylene chloride:hexanes, and compound AP-7 was isolated in 16% yield.

Example 4
Selective Oxidation of 4-amino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one (AP-2) to 2,3-dimethyl-4-hydroxyamino-1-phenyl-3-pyrazolin-5-one (AP-3)

4-Aminoantipyrine (0.12 mmol, Aldrich) and octachloro-octachloro tetrasulfonato Fe(III) porphyrin (0.6 μmol) were dissolved in 4 mL of pH 7 phosphate buffer and cooled to 0°C. To this was added iodosobenzene (66 mg, 0.3 mmol) was added in portions over 10 min. The reaction was stirred for 4 hr and extracted with 3 x 2 mL of methylene chloride and dried over Na_2CO_3. The organic layer was chromatographed over silica gel, eluting with methylene chloride, to yield 2,3-dimethyl-4-hydroxyamino-1-phenyl-3-pyrazolin-5-one (AP-3) in 30 % yield.

Example 5
Selective oxidation of 4-acetylaminoantipyrine (AP-8)

Cumene hydroperoxide (150 mg, 0.99 mmol) was added slowly to a solution of 4-acetylaminoantipyrine (120 mg, 0.49 mmol) and octachloro-octachloro tetrasulfonato Fe(III) porphyrin (15 mg, 0.95 μmol) held at 5°C, and the mixture was allowed to stand for 3 hr at room temperature. The mixture was made weakly basic with aqueous Na_2CO_3, and the product was extracted with 3 x 5 mL of methylene chloride. The organic extract was dried over Na_2CO_3 and evaporated under reduced pressure. The residue was chromatographed over basic alumina, eluting with 1:1 methylene chloride:hexane. The fraction containing the last band to come off the column was collected and evaporated under reduced pressure. The residue was crystallized from methylene chloride:hexane to obtain the compound AP-9 (see Scheme 3) in 32% yield.

Example 6
Selective oxidation of
3-hydroxymethylaminopyrine (AP-5)

Following the procedure of Example 2 above, 4-dimethylamino-3-hydroxymethyl-2-methyl-1-phenyl-3-pyrazolin-5-one (AP-5) was substituted for the aminopyrine thereof. After 2 hr of reaction at 5°C, the mixture was neutralized with 0.1 N NaOH. The products were extracted with 3 x 5 mL of methylene chloride. The organic extract was dried over Na_2CO_3 and evaporated under reduced pressure. The residue was chromatographed over neutral alumina, eluting with methylene chloride, to obtain the 3-formyl derivative in 45 % yield (AP-10, Scheme 4).

Example 7
Oxidation of lidocaine

Six mg (3.8 µmol) of octachloro-octachloro tetrasulfonato Fe(III) porphyrin and 714 (3 mmol, Aldrich) of lidocaine were dissolved in a 1:2 acetonitrile:pH7 phosphate buffer. To this solution was added 1.7 g (8 mmol) of iodosobenzene, and the reaction was stirred for 4 hr at room temperature. The solution was extracted with 3 x 5 mL of methylene chloride. The organic extract was dried over $MgSO_4$ and evaporated under reduced pressure. The residue was chromatographed over neutral alumina, eluting with 1:1 methylene chloride:hexane. Six metabolites were obtained (see Scheme 5): ω-(ethylamino)-2,6-dimethylacetanilide (L-1) 34%, ω-amino-2,6-dimethylacetanilide (L-2) 6%, ω-diethylamino-2-hydroxymethyl-6-methylacetanilide (L-3) 4 %, and lidocaine N-oxide (L-4) 15%, and small amounts of cyclic metabolites, 1-ethyl-2-methyl-3-(2,6-dimethylphenyl)imidazolin-4-one (L-8) 2%, and 2-methyl-3-(2,6-dimethylphenyl)-imidazolin-4-one (L-9) 3%.

Example 8
Oxidation of lidocaine
in organic solvents

Iodosobenzene (0.3 mmol) was progressively added over 30 min at 0°C to a 2 mL solution of lidocaine HCl (0.15 mmol) and of octachloro-octachloro tetrasulfonato Fe(III) porphyrin (0.5 µmol) in 1:20 methanol:acetonitrile. After 5 hr, the solution was evaporated at 20°C under reduced pressure. The residue was chromatographed over neutral alumina, eluting with benzene. Compound L-1

and L-2 w re obtained as major products and L-8 as a minor product; compounds which had not previously been reported to be metabolites of lidocaine include 3-hydroxy-ω-diethylamino-2,6-dimethylacetanilide (L-5) 13%, and 4-hydroxy-ω-diethylamino-2,6-dimethylacetanilide (L-6) 2%, 2-hydroxymethyl-ω-diethylamino-2,6-dimethylacetanilide (L-7) 2%, as well as a small amounts of L-4 and L-9 (all known compounds).

Example 9
Oxidation of 2,6-dimethylaniline

Example 8 above was repeated, substituting 2,6-dimethylaniline (L-7, see Scheme 6) for the lidocaine thereof. Oxidation products which were isolated are: 2,6-dimethylphenylhydroxylamine (L-7) 12%, 4-hydroxy-2,6-dimethylaniline (L-8) 5%, and 2,6-dimethylbenzoquinone (L-9) 2%.

Example 10
Preparation of octachloro-octabromoporphyrinato-iron(III)

10a. Octachloroporphyrinato-zinc(II)

2,6-Dichlorobenzaldehyde (100 g, 0.57 mol), anhydrous zinc acetate (30 g), and 2,6-lutidine (300 mL) were heated in a 1 L 3-neck flask fitted with a reflux condenser and a drying tube. When the temperature reached 100°C, pyrrole (40 mL, 0.57 mol) was added dropwise within 10 min, and the reaction mixture was refluxed for 16 hr. The solvent was removed under vacuum, and the residue was triturated with toluene (400 mL). Methanol (100 mL) was added and the mixture was held at 5°C for 16 hr. The precipitate was collected by filtration and dissolved in 500 mL of chlorform, to which was then added 50 mL of trifluoroacetic acid. The mixture was stirred at room temperature for 16 hr under nitrogen. Water (250 mL) was added, and the mixture was stirred vigorously for 10 min. The organic phase was washed with satd $NaHCO_3$ and water, dried over Na_2SO_4. Ten g of p-chloranil was added, and the mixture was refluxed for 2 hr under N_2. The mixture was held at room temperature for 16 hr, reheated to reflux, and clarified by passage through a column of alumina. The solution was concentrated to a 200 mL volume, and 200 mL of methanol was added. The chloroform was removed from the solvent under vacuum, and the resulting suspension was filtered. The porphyrin (27 g) was dissolved in 500 mL of DMF, solid zinc acetate was added, and the mixture was refluxed for 30 min. The resulting precipitate was collected, washed with water and methanol, then dried

to give 7.26 g of the title compound as a purple solid. UV/vis absorption (CH_2Cl_2), nm (relative intensity): 627.0 (0.91), 584.5 (1.71, (550.0 (12.08), 513.0 (1.82), 486.5 (1.49), 420.0 (100.0), 399.0 (23.1). MS m/z:1585 $(M+H)^+$.

10b. Alternate, large-scale preparation of octachloroporphyrinato-zinc(II)

2,6-Dichlorobenzaldehyde (750 g, 4.29 mol), anhydrous zinc acetate (580 g), and 2,6-lutidine (5.0 L) were heated in a 12 L 4-neck flask fitted with a reflux condenser and a drying tube. When the temperature reached 90-100°C, pyrrole (600 mL, 4.29 mol) was added slowly within 10 min, and the reaction mixture was refluxed for 18 hr. The solvent was removed under vacuum, and the residue was triturated with toluene (6 L). Methanol (500 mL) was added and the mixture was held at 5°C for 16 hr. The precipitate was collected by filtration, rinsed with methanol, and dried under vacuum. The dried compound was suspended in 6.0 L of hot chloroform and 6.0 L of methanol was added. The solvent was slowly evaporated under vacuum to remove the chloroform, and the resulting purple precipitate was collected by filtration, rinsed with methanol, water and pentane, and dried under vacuum to give 357 g of the title compound. A 30 g sample of this crude product was dissolved in 1.5 L of chloroform and 150 mL of Trifluoroacetic acid was added slowly. The mixture was stirred at room temperature for 16 hr under N_2. Water (1.5 L) was added, and the mixture was stirred vigorously for 10 min. The organic phase was washed with satd $NaHCO_3$ and water, dried over Na_2SO_4, transferred to a round-bottom flask, then 30 g of p-chloranil was added and the mixture was refluxed for 3 hr under N_2. The hot solution was passed through a column of alumina, which was rinsed with hot chloroform. The solution was concentrated to a 200 mL volume, and 200 mL of methanol was added. The chloroform was removed from the under vacuum, and the resulting suspension was filtered, to yield 7.98 g of the porphyrin. The porphyrin was dissolved in 750 mL of DMF, solid zinc acetate (20 g) was added, and the mixture was refluxed for 2 hr. The solution was cooled, 550 mL of DMF was distilled off under vacuum, and 200 mL of water was added. The resulting precipitate was collected, washed with water and methanol, then dried to give 9.13 g of the title compound as a purple solid.

10c. Octachloro-octabromoporphyrinato-zinc(II)

A 0.5 g (0.525 mmol) sample of the octachloro zinc porphyrin compound (from step 11a or 11b above) was dissolved in 50 mL of methanol and treated with bromine (0.27 mL, 5.25 mmol). The resulting mixture was heated at reflux for 1 hr. The mixture was taken to dryness, and the residue was chromatographed on alumina (neutral, Brockmann type 1), eluting with $CHCl_3$. The dark green band was collected and the solvent was removed under vacuum to afford the title

compound as a green solid (0.332 g, 39% yield). UV/vis absorption (CH_2Cl_2), nm (relative intensity): 594 (7.49), 462 (100), 371 (14.15).

10d. Octachloro-octabromoporphyrinato-iron(III)

The compound from the step 11c was convert d into the hemin metalloporphyrin by the method of Kobayashi *et al.* (*Bull. Chem. Soc. Japan*, 48:3137 (1975)).

Example 11

Alternate preparation of octachloro-octabromoporphyrinato-iron(III)

11a. Octachloro-octabromoporphyrinato-zinc(II)

A sample of *meso*-tetrakis-(2,6-dichlorophenyl)porphyrinato-zinc(II) (0.5 g, 0.525 mmol, from steps 10a or 10b) was dissolved in 50 mL of methanol and treated with bromine (0.27 mL, 5.25 mmol). The resulting mixture was stirred at room temperature for 1.5 hr, and held at 4°C for 16 hr. The precipitate was collected by filtration and washed with a small amount of methanol to give 0.268 g (65% yield) of the title product. UV-vis absorption (CH_2Cl_2), nm (relative intensity): 594 (7.0), 463 (100), 368.5 (11.5). MS M/Z $(m+H)^+$: 1585.

11b. Octachloro-octabromoporphyrinato-iron(III)

The compound from the step 11a was converted into the hemin metalloporphyrin by the method of Kobayashi *et al.* (*Bull. Chem. Soc. Japan*, 48:3137 (1975)).

Example 12

Preparation of octachloro-octachloroporphyrinato-iron(III)

12a. Octachloro-octachloroporphyrinato-zinc(II)

A sample of *meso*-tetrakis-(2,6-dichlorophenyl)porphyrinato-zinc(II) (0.250 g, 0.262 mmol, from steps 10a or 10b) was suspended in 50 mL of methanol, and the mixture was cooled to 0°C with an ice bath. Chlorine gas was bubbled into the mixture at a rate such that the temperature remained below 5°C. After 15-20 minutes, the color of the reaction mixture changed from purple to green, and starting material was no longer present. The solvent and unreacted chlorine was removed under vacuum to give a purple solid (0.283 g 88% yield). UV-vis absorption (CH_2Cl_2), nm (relative intensity): 436.0 (100), 485.0 (50), 523.0 (9.5), 575.0 (9.5), 625.0 (9.3). MS M/Z (relative intensity): 1338 (25), 1305 (45), 1266 (50), 1230 (100), 1195 (60), 1160 (55).

12b. Octachloro-octachloroporphyrinato-iron(III)

The compound from the step 12a was converted into th hemin metalloporphyrin by the method of Kobayashi *et al.* (*Bull. Chem. Soc. Japan*, 48:3137 (**1975**)).

Example 13
Oxidation of ABT-418 in organic solvent

ABT-418 (0.506 mg) was dissolved in 10 mL of methylene chloride, and 8 mg octachloro-octabromo Fe(III) porphyrin (0.5 µmol) was added. The mixture was stirred vigorously, and 0.511 mg of iosdosobenzene was added in small portions . After 5 hr, the solution was evaporated at 20°C under reduced pressure, and the residue was redissolved in 10 mL of methylene chloride. The solution was analyzed by HPLC, and the products were identified by comparison with known samples of metabolites (cf., Sullivan *et al.*, mss in preparation).

Example 14
Oxidation of ABT-418 in aqueous system

ABT-418 (0.498 g) was dissolved in 10 mL of 4:1 acetonitrile:water solution . To this was added 8 mg of octachloro-octabromo-tetrasulfonate Fe(III) porphyrin, followed by 2 mL of aqueous sodium hypochlorite. The reaction was stirred for 8 hr, and the layers were separated. The layers were examined by HPLC, and the products were identified by comparison with known samples of metabolites.

Example 15
Oxidation of odapipam

Odapipam (10 µmol) was dissolved in 1 mL of methylene chloride, and 1 µmol of pentafluoroFe(III) porphyrin and 40 µmol of iodosobenzene were added. After reaction was complete, the products were separated by HPLC and identified by mass spectroscopy (cf O'Boyle *et al.*, *Pharmac. Th rap.*, 43:1, (**1989**)).

Example 16
Systematic oxidation of aminopyrine

Twenty-seven flasks, each containing 50 mg of aminopyrine are prepared, and solvents, synthetic metalloporphyrins and oxidizing reagents are added to the flasks according to Table 4a-4c below (see abbreviations below). The contents are mixed for 4 hr at room temperature, and the solvents are removed under vacuum. The residues are taken up in methylene chloride, and the products are identified by HPLC, mass spectroscopy, NMR spectroscopy and elemental analysis.

Abbreviations used in Tables 4a-4c below: A= methylene chloride; B= 20% (v/v) acetonitrile in water; C= pH 6 phosphate buffer; D= octachloro-octabromo Fe(III) porphyrin; E= octachloro-octabromo MN(II) porphyrin; F= octachloro-octachloro tetrasulfonato Fe(III) porphyrin; G=octachloro-octachloro tetrasulfonato Mn(II) porphyrin; H=iodosobenzene; I=sodium hypochlorite; J=*tert*-butyl hydroperoxide; and K=potassium monopersulfate.

TABLE 4a. Combination of solvent, smp and oxidizing reagents in flasks 1-16.

Fl	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
S	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
smp	D	D	D	D	E	E	E	E	F	F	F	F	G	G	G	G
ox	H	I	J	K	H	I	J	K	H	I	J	K	H	I	J	K

Fl=flask; S=solvent; smp =synthetic metalloporphyrin; ox=SMP-co-oxidizing reagents

TABLE 4b. Combination of solvent, smp and oxidizing reagents in flasks 17-32.

Fl	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32
S	B	B	B	B	B	B	B	B	B	B	B	B	B	B	B	B
smp	D	D	D	D	E	E	E	E	F	F	F	F	G	G	G	G
ox	H	I	J	K	H	I	J	K	H	I	J	K	H	I	J	K

Fl=flask; S=solvent; smp =synthetic metalloporphyrin; ox=SMP-co-oxidizing r agents

TABLE 4c. Combination of solvent, smp and oxidizing reagents in flasks 32-48.

Fl	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48
S	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C	C
smp	D	D	D	D	E	E	E	E	F	F	F	F	G	G	G	G
ox	H	I	J	K	H	I	J	K	H	I	J	K	H	I	J	K

Fl=flask; S=solvent; smp =synthetic metalloporphyrin; ox=SMP-co-oxidizing reagents

CLAIMS

What is claimed is:

1. A process for the systematic preparation of oxidative products of a drug candidate compound, comprising reacting samples of the drug candidate compound with a series of combinations of a selected synthetic metalloporphyrin with a selected SMP-co-oxidizing reagent in the presence of a suitable solvent, for a period of up to 24 hours, at a temperature from 0°C to reflux temperature of the solvent, in a manner such that each sample of drug compound is reacted with a different combination of synthetic metalloporphyrin, SMP-co-oxidizing reagent and solvent, then separating and isolating the resulting oxidative products.

2. The process according to Claim 1, wherein the selected synthetic metalloporphyrins are from the group consisting of octachloro-octabromo Fe(III) porphyrin, octachloro-octabromo Mn(II) porphyrin, octachloro-octachloro Fe(III) porphyrin, octachloro-octachloro Mn(II) porphyrin, octachloro-octabromo tetrasulfonato Fe(III) porphyrin, octachloro-octabromo tetrasulfonato Mn(II) porphyrin, octachloro-octachloro tetrasulfonato Fe(III) porphyrin, octachloro-octachloro tetrasulfonato Mn(II) porphyrin, octachloro-tetranitro Fe(III) porphyrin, octachloro-tetranitro Mn(II) porphyrin, octachloro-octacyano Fe(III) porphyrin, and octachloro-octacyano Mn(II) porphyrin.

3. The process according to Claim 1, wherein the selected SMP-co-oxidizing reagents are from the group consisting of iodosobenzene, sodium hypochlorite, potassium monopersulfate, ozone, hydrogen peroxide, m-chloroperbenzoic acid, cumene hydroperoxide and *tert*-butyl hydroperoxide.

4. The process according to Claim 1, wherein the solvents are selected from the group consisting of CH_2Cl_2, CH_3CN, 20% methanol in H_2O, 20% CH_3CN in H_2O, and buffered aqueous solutions thereof.

5. The process according to Claim 1, wherein the synthetic metalloporphyrins are selected from the group consisting of octachloro-octabromo Fe(III) porphyrin, octachloro-octabromo Mn(II) porphyrin, octachloro-octachloro tetrasulfonato F (III) porphyrin, and octachloro-octachloro tetrasulfonato Mn(II) porphyrin; the SMP-co-oxidizing reagents are selected from the group consisting of iodosobenzene, sodium hypochlorite, *tert*-butyl hydroperoxide, and potassium monopersulfate; and the solvents are those selected from the group consisting of CH_2Cl_2, 20% CH_3CN in H_2O, and buffered aqueous solutions thereof.

6. A process for the preparation, identification and further testing of pharmaceutical product metabolites comprising systematically preparing oxidative products of a pharmaceutical product according to the process of Claim 1, followed by confirming the identities of said metabolites in appropriate animal model studies using the oxidative products as a guide, and further testing of these metabolites in toxicologic, pathologic, histopathologic, mechanistic or genotoxic studies in order to identify toxic or otherwise biologically-active metabolites of the pharmaceutical product.

PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : C07B 33/00, B01J 31/18 — A3

(11) International Publication Number: WO 96/08455

(43) International Publication Date: 21 March 1996 (21.03.96)

(21) International Application Number: PCT/US95/11522

(22) International Filing Date: 14 September 1995 (14.09.95)

(30) Priority Data:

08/306,801	15 September 1994 (15.09.94)	US
08/520,842	12 September 1995 (12.09.95)	US

(71) Applicant: ABBOTT LABORATORIES [US/US]; CHAD 0377/AP6D-2, 100 Abbott Park Road, Abbott Park, IL 60064-3500 (US).

(72) Inventors: CHORGHADE, Mukund, S.; 5048 Adele Drive, Gurnee, IL 60031 (US). DOLPHIN, David, H.; 4464 West 12th Avenue, Vancouver, British Columbia V6R 2R2 (CA). HILL, David, R.; Apartment E, 5312 George Court, Gurnee, IL 60031 (US). HINO, Fumio; 1-35-23, Noyawa, Setagaya-ku, Tokyo 154 (JP). LEE, Elaine, C.; 755-B Brookvale Drive, Wheeling, IL 60090 (US).

(74) Agents: ELDER, Richard, A. et al.; Abbott Laboratories, CHAD 0377/AP6D-2, 100 Abbott Park Road, Abbott Park, IL 60064-3500 (US).

(81) Designated States: CA, JP, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(88) Date of publication of the international search report: 13 June 1996 (13.06.96)

(54) Title: USE OF SYNTHETIC METALLOPORPHYRINS FOR PREPARATION AND PREDICTION OF DRUG METABOLITES

(57) Abstract

A method for the systematic and efficient synthetic preparation and identification of metabolites of a pharmaceutical product in order to study possible toxic and/or otherwise biologically-active metabolites of such pharmaceutical products as early and conveniently as possible in the very expensive drug development process, comprising adding samples of the pharmaceutical product to a series of combinations of a synthetic metalloporphyrin (SMP) with a synthetic metalloporphyrin-co-oxidizing reagent in the presence of a suitable solvent, under specified conditions, in a manner such that each sample of pharmaceutical product is reacted with a different combination of synthetic metalloporphyrin, SMP-co-oxidizing reagent and solvent, followed by separation and isolation of the resulting oxidative products, then confirmation of the identity of metabolites from the pre-identified oxidative products by appropriate animal model studies, and subjecting the actual metabolites prepared in larger quantities by the above method to toxicologic, pathologic, histopathologic, mechanistic or genotoxic testing in order to identify toxic and/or otherwise metabolically-active beneficial or detrimental individual metabolites.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AT	Austria	**GB**	United Kingdom	**MR**	Mauritania
AU	Australia	**GE**	Georgia	**MW**	Malawi
BB	Barbados	**GN**	Guinea	**NE**	Niger
BE	Belgium	**GR**	Greece	**NL**	Netherlands
BF	Burkina Faso	**HU**	Hungary	**NO**	Norway
BG	Bulgaria	**IE**	Ireland	**NZ**	New Zealand
BJ	Benin	**IT**	Italy	**PL**	Poland
BR	Brazil	**JP**	Japan	**PT**	Portugal
BY	Belarus	**KE**	Kenya	**RO**	Romania
CA	Canada	**KG**	Kyrgystan	**RU**	Russian Federation
CF	Central African Republic	**KP**	Democratic People's Republic of Korea	**SD**	Sudan
CG	Congo			**SE**	Sweden
CH	Switzerland	**KR**	Republic of Korea	**SI**	Slovenia
CI	Côte d'Ivoire	**KZ**	Kazakhstan	**SK**	Slovakia
CM	Cameroon	**LI**	Liechtenstein	**SN**	Senegal
CN	China	**LK**	Sri Lanka	**TD**	Chad
CS	Czechoslovakia	**LU**	Luxembourg	**TG**	Togo
CZ	Czech Republic	**LV**	Latvia	**TJ**	Tajikistan
DE	Germany	**MC**	Monaco	**TT**	Trinidad and Tobago
DK	Denmark	**MD**	Republic of Moldova	**UA**	Ukraine
ES	Spain	**MG**	Madagascar	**US**	United States of America
FI	Finland	**ML**	Mali	**UZ**	Uzbekistan
FR	France	**MN**	Mongolia	**VN**	Viet Nam
GA	Gabon				

INTERNATIONAL SEARCH REPORT

Inter .al Application No
PCT/US 95/11522

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 C07B33/00 B01J31/18

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 C07B B01J

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO,A,88 07988 (DOLPHIN ET AL.) 20 October 1988 see page 2, line 1 - page 2, line 12 ---	1-6
P,Y	EP,A,0 636 628 (SUN COMPANY, INC. (R & M)) 1 February 1995 see page 3, line 55 - page 3, line 56 ---	1-6
Y	TETRAHEDRON LETT., vol. 30, no. 39, 1989 pages 5231-4, J. A. SHELNUTT, D. E. TRUDELL 'PHOTOCHEMICALLY-DRIVEN BIOMIMETIC OXIDATION OF ALKANES AND OLEFINS' * entire document * --- -/--	1-6

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

1

Date of the actual completion of the international search	Date of mailing of the international search report
11 March 1996	03.05.96

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+ 31-70) 340-2040, Tx. 31 651 epo nl, Fax (+ 31-70) 340-3016	Herz, C

Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	BULL. SOC. BELG., vol. 95, no. 11, 1986 pages 959-964, D. MANSUY, P. BATTIONI 'BIOMIMETIC OXIDATION OF HYDROCARBONS AND DRUGS BY METALLOPORPHYRINIC SYSTEMS' * entire document * ---	1-6
X	BULL. SOC. CHIM. FR., vol. 131, no. 6, 1994 pages 706-712, N. GAGGERO ET AL. 'Oxidation of SR 48117, an antagonist of vasopressin V1a receptors, by biomimetic catalysts based on metalloporphyrin or Schiff-base complexes' * entire document * ---	1-6
X	BULL. SOC. CHIM. FR., vol. 130, no. 3, 1993 pages 405-416, M. N. CARRIER ET AL. 'Studying drug metabolic oxidation with biomimetic metalloporphyrin systems: Problems and solutions in the case of lidocaine' * entire document * ---	1-6
X	DRUG MET. DISPOS., vol. 21, no. 5, 1993 pages 811-817, M. VIDAL ET AL. 'MODEL SYSTEMS FOR OXIDATIVE DRUG METABOLISM STUDIES: Catalytic Behaviour of Water-Soluble Metalloporphyrins Depends on Both the Intrinsic Robustness of the Catalyst and the Nature of the Substrates' * entire document * ---	1-6
X	DRUG MET. DISP., vol. 19, no. 2, 1991 pages 360-365, J. BERNADOU ET AL. 'MODEL SYSTEMS FOR METABOLISM STUDIES: Biomimetic Oxidation of Acetaminophen and Ellipticine Derivatives with Water-Soluble Metalloporphyrins Associated to Potassium Monopersulfate' * entire document * --- -/--	1-6

1

INTERNATIONAL SEARCH REPORT

Inter. nal Application No
PCT/US 95/11522

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	J. MOL. CATAL., vol. 29, no. 2, 1985 pages 153-156, H. SAKURAI ET AL. 'A Model System for Drug Metabolism in Isolated Hepatocytes; Oxidation of Cyclohexene by Metalloporphyrin Complexes' * entire document * -----	1-6

1

Form PCT/ISA/210 (continuation of second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Information on patent family members

Inter. nal Application No
PCT/US 95/11522

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
WO-A-8807988	20-10-88	AT-T-	123749	15-06-95
		AU-B-	617670	05-12-91
		AU-B-	1707588	04-11-88
		CA-A-	1308096	29-09-92
		DE-D-	3853995	20-07-95
		DE-T-	3853995	23-11-95
		EP-A-	0363379	18-04-90
		FI-B-	92402	29-07-94
		JP-T-	2503086	27-09-90
		US-A-	4892941	09-01-90
		US-A-	5077394	31-12-91
EP-A-636628	01-02-95	US-A-	5480986	02-01-96
		CA-A-	2129055	31-01-95
		JP-A-	7089964	04-04-95